

# Dielectric Relaxation of Hydration Water in Native Collagen Fibrils

Kurzweil-Segev Y., Popov I., Solomonov I., Sagit I., Feldman Y.  
*Kazan Federal University, 420008, Kremlevskaya 18, Kazan, Russia*

---

## Abstract

© 2017 American Chemical Society. The dielectric relaxation of hydrated collagen powders was studied over a wide temperature and frequency range. We revealed two mechanisms of dielectric relaxation in hydration water that are driven by the migration of ionic and orientation defects. At high water fractions in powders ( $h > 0.2$ ), the hydration shell around the collagen triple helix presents a spatial H-bonded network consisting of structural water bridges and cleft water channels. These two water phases provide the long-range paths for proton hopping and orientation defect migration. At low water fractions ( $h < 0.2$ ) and in the hydrated collagen samples after the dehydrothermal treatment, the hydration shell presents localized individual water compartments not connected to one another. In these cases, the relaxation mechanism due to proton hopping either disappears or becomes inhibited by the orientation defect migration.

<http://dx.doi.org/10.1021/acs.jpcb.7b02404>

---

## References

- [1] Busch, S.; Bruce, C. D.; Redfield, C.; Lorenz, C. D.; McLain, S. E. Water Mediation Is Essential to Nucleation of beta-Turn Formation in Peptide Folding Motifs *Angew. Chem., Int. Ed.* 2013, 52, 13091-13095 10.1002/anie.201307657
- [2] Chaplin, M. Opinion-Do We Underestimate the Importance of Water in Cell Biology? *Nat. Rev. Mol. Cell Biol.* 2006, 7, 861-866 10.1038/nrm2021
- [3] Ladbury, J. E. Just Add Water! The Effect of Water on the Specificity of Protein-Ligand Binding Sites and Its Potential Application to Drug Design *Chem. Biol.* 1996, 3, 973-980 10.1016/S1074-5521(96)90164-7
- [4] Levy, Y.; Onuchic, J. N. Water and Proteins: A love-Hate Relationship *Proc. Natl. Acad. Sci. U.S.A.* 2004, 101, 3325-3326 10.1073/pnas.0400157101
- [5] Levy, Y.; Onuchic, J. N. Water Mediation in Protein Folding and Molecular Recognition *Annu. Rev. Biophys. Biomol. Struct.* 2006, 35, 389-415 10.1146/annurev.biophys.35.040405.102134
- [6] Meyer, E. Internal Water-Molecules and H-Bonding in Biological Macromolecules-a Review of Structural Features with Functional Implications *Protein Sci.* 1992, 1, 1543-1562 10.1002/pro.5560011203
- [7] Tame, J. R. H.; Sleight, S. H.; Wilkinson, A. J.; Ladbury, J. E. The Role of Water in Sequence-Independent Ligand Binding by an Oligopeptide Transporter Protein *Nat. Struct. Biol.* 1996, 3, 998-1001 10.1038/nsb1296-998
- [8] Dielmann-Gessner, J.; Grossman, M.; Nibali, V. C.; Born, B.; Solomonov, I.; Fields, G. B.; Havenith, M.; Sagi, I. Enzymatic Turnover of Macromolecules Generates Long-Lasting Protein-Water-Coupled Motions Beyond Reaction Steady State *Proc. Natl. Acad. Sci. U.S.A.* 2014, 111, 17857-17862 10.1073/pnas.1410144111
- [9] Grossman, M.; Born, B.; Heyden, M.; Tworowski, D.; Fields, G. B.; Sagi, I.; Havenith, M. Correlated Structural Kinetics and Retarded Solvent Dynamics at the Metalloprotease Active Site *Nat. Struct. Mol. Biol.* 2011, 18, 1102-U1113 10.1038/nsmb.2120

- [10] Algara-Siller, G.; Lehtinen, O.; Wang, F. C.; Nair, R. R.; Kaiser, U.; Wu, H. A.; Geim, A. K.; Grigorieva, I. V. Square Ice in Graphene Nanocapillaries *Nature* 2015, 519, 443-445 10.1038/nature14295
- [11] Kou, J. L.; Lu, H. J.; Wu, F. M.; Fan, J. T.; Yao, J. Electricity Resonance-Induced Fast Transport of Water through Nanochannels *Nano Lett* 2014, 14, 4931-4936 10.1021/nl500664y
- [12] Moore, E. B.; Allen, J. T.; Molinero, V. Liquid-Ice Coexistence Below the Melting Temperature for Water Confined in Hydrophilic and Hydrophobic Nanopores *J. Phys. Chem. C* 2012, 116, 7507-7514 10.1021/jp3012409
- [13] Ebbinghaus, S.; Kim, S. J.; Heyden, M.; Yu, X.; Heugen, U.; Gruebele, M.; Leitner, D. M.; Havenith, M. An Extended Dynamical Hydration Shell Around Proteins *Proc. Natl. Acad. Sci. U.S.A.* 2007, 104, 20749-20752 10.1073/pnas.0709207104
- [14] Mathur-De Vré, R. The NMR Studies of Water in Biological Systems *Prog. Biophys. Mol. Biol.* 1979, 35, 103-134 10.1016/0079-6107(80)90004-8
- [15] Nittinger, E.; Schneider, N.; Lange, G.; Rarey, M. Evidence of Water Molecules-A Statistical Evaluation of Water Molecules Based on Electron Density *J. Chem. Inf. Model* 2015, 55, 771-783 10.1021/ci500662d
- [16] Rhee, Y. M.; Sorin, E. J.; Jayachandran, G.; Lindahl, E.; Pande, V. S. Simulations of the Role of Water in the Protein-Folding Mechanism *Proc. Natl. Acad. Sci. U.S.A.* 2004, 101, 6456-6461 10.1073/pnas.0307898101
- [17] Zhang, L. Y.; Wang, L. J.; Kao, Y. T.; Qiu, W. H.; Yang, Y.; Okobiah, O.; Zhong, D. P. Mapping Hydration Dynamics around a Protein Surface *Proc. Natl. Acad. Sci. U.S.A.* 2007, 104, 18461-18466 10.1073/pnas.0707647104
- [18] Fassett, J.; Tobolt, D.; Hansen, L. K. Type I Collagen Structure Regulates Cell Morphology and EGF Signaling in Primary Rat Hepatocytes Through cAMP-Dependent Protein Kinase A *Mol. Biol. Cell* 2006, 17, 345-356 10.1091/mbc.E05-09-0871
- [19] Hansen, L. K.; Wilhelm, J.; Fassett, J. T. Regulation of Hepatocyte Cell Cycle Progression and Differentiation by Type I Collagen Structure *Curr. Top. Dev. Biol.* 2006, 72, 205-236 10.1016/S0070-2153(05)72004-4
- [20] Huang, Y.; Arora, P.; McCulloch, C. A.; Vogel, W. F. The Collagen Receptor DDR1 Regulates Cell Spreading and Motility by Associating With Myosin IIA *J. Cell Sci.* 2009, 122, 1637-1646 10.1242/jcs.046219
- [21] Koohestani, F.; Braundmeier, A. G.; Mahdian, A.; Seo, J.; Bi, J. J.; Nowak, R. A. Extracellular Matrix Collagen Alters Cell Proliferation and Cell Cycle Progression of Human Uterine Leiomyoma Smooth Muscle Cells *PLoS One* 2013, 8 e75844 10.1371/journal.pone.0075844
- [22] Rasmussen, C. H.; Petersen, D. R.; Moeller, J. B.; Hansson, M.; Dufva, M. Collagen Type I Improves the Differentiation of Human Embryonic Stem Cells towards Definitive Endoderm *PLoS One* 2015, 10 e0145389 10.1371/journal.pone.0145389
- [23] Solomonov, I.; Zehorai, E.; Talmi-Frank, D.; Wolf, S. G.; Shainskaya, A.; Zhuravlev, A.; Kartvelishvili, E.; Visse, R.; Levin, Y.; Kampf, N. et al. Distinct Biological Events Generated by ECM Proteolysis by Two Homologous Collagenases *Proc. Natl. Acad. Sci. U.S.A.* 2016, 113, 10884-10889 10.1073/pnas.1519676113
- [24] Bella, J.; Eaton, M.; Brodsky, B.; Berman, H. M. Crystal-Structure and Molecular-Structure of a Collagen-Like Peptide at 1.9-Angstrom Resolution *Science* 1994, 266, 75-81 10.1126/science.7695699
- [25] Vanderrest, M.; Garrone, R. Collagen Family of Proteins *FASEB J.* 1991, 5, 2814-2823
- [26] Ramachandran, G. N.; Chandrasekharan, R. Interchain Hydrogen Bonds Via Bound Water Molecules in Collagen Triple Helix *Biopolymers* 1968, 6, 1649-1658 10.1002/bip.1968.360061109
- [27] Ramachandran, G. N. *Treatise on Collagen*; Academic Press: New York, 1967.
- [28] Bella, J.; Brodsky, B.; Berman, H. M. Hydration Structure of a Collagen Peptide Structure 1995, 3, 893-906 10.1016/S0969-2126(01)00224-6
- [29] Fullerton, G. D.; Nes, E.; Amurao, M.; Rahal, A.; Krasnosselskaia, L.; Cameron, I. An NMR Method to Characterize Multiple Water Compartments on Mammalian Collagen *Cell Biol. Int.* 2006, 30, 66-73 10.1016/j.cellbi.2005.09.009
- [30] Mogilner, I. G.; Ruderman, G.; Grigera, J. R. Collagen Stability, Hydration and Native State *J. Mol. Graphics Modell.* 2002, 21, 209-213 10.1016/S1093-3263(02)00145-6
- [31] Berendsen, H. J. C.; Migchelsen, C. Hydration Structure of Fibrous Macromolecules *Ann. N. Y. Acad. Sci.* 1965, 125, 365-379 10.1111/j.1749-6632.1965.tb45403.x
- [32] Grigera, J. R.; Berendsen, H. J. C. Molecular Details of Collagen Hydration *Biopolymers* 1979, 18, 47-57 10.1002/bip.1979.360180106
- [33] Peto, S.; Gillis, P.; Henri, V. P. Structure and Dynamics of Water in Tendon from NMR Relaxation Measurements *Biophys. J.* 1990, 57, 71-84 10.1016/S0006-3495(90)82508-X
- [34] Fullerton, G. D.; Amurao, M. R. Evidence that Collagen and Tendon Have Monolayer Water Coverage in the Native State *Cell Biol. Int.* 2006, 30, 56-65 10.1016/j.cellbi.2005.09.008
- [35] Berendsen, H. J. C. Nuclear Magnetic Resonance Study of Collagen Hydration *J. Chem. Phys.* 1962, 36, 3297-3305 10.1063/1.1732460
- [36] Gainaru, C.; Fillmer, A.; Bohmer, R. Dielectric Response of Deeply Supercooled Hydration Water in the Connective Tissue Proteins Collagen and Elastin *J. Phys. Chem. B* 2009, 113, 12628-12631 10.1021/jp9065899

- [37] Lusceac, S. A.; Rosenstihl, M.; Vogel, M.; Gainaru, C.; Fillmer, A.; Bohmer, R. NMR and Dielectric Studies of Hydrated Collagen and Elastin: Evidence for a Delocalized Secondary Relaxation J. Non-Cryst. Solids 2011, 357, 655-663 10.1016/j.jnoncrysol.2010.07.035
- [38] Lusceac, S. A.; Vogel, M. R.; Herbers, C. R. H-2 and C-13 NMR Studies on the Temperature-Dependent Water and Protein Dynamics in Hydrated Elastin, Myoglobin and Collagen Biochim. Biophys. Acta, Proteins Proteomics 2010, 1804, 41-48 10.1016/j.bbapap.2009.06.009
- [39] Vogel, M. Origins of Apparent Fragile-to-Strong Transitions of Protein Hydration Waters Phys. Rev. Lett. 2008, 101 225701 10.1103/PhysRevLett.101.225701
- [40] Wess, T. J.; Orgel, J. P. Changes in Collagen Structure: Drying, Dehydrothermal Treatment and Relation to Long Term Deterioration Thermochim. Acta 2000, 365, 119-128 10.1016/S0040-6031(00)00619-5
- [41] Jansson, H.; Swenson, J. The Protein Glass Transition as Measured by Dielectric Spectroscopy and Differential Scanning Calorimetry Biochim. Biophys. Acta, Proteins Proteomics 2010, 1804, 20-26 10.1016/j.bbapap.2009.06.026
- [42] Khodadadi, S.; Pawlus, S.; Roh, J. H.; Sakai, V. G.; Mamontov, E.; Sokolov, A. P. The Origin of the Dynamic Transition in Proteins J. Chem. Phys. 2008, 128 195106 10.1063/1.2927871
- [43] Khodadadi, S.; Pawlus, S.; Sokolov, A. P. Influence of Hydration on Protein Dynamics: Combining Dielectric and Neutron Scattering Spectroscopy Data J. Phys. Chem. B 2008, 112, 14273-14280 10.1021/jp8059807
- [44] Nakanishi, M.; Sokolov, A. P. Protein Dynamics in a Broad Frequency Range: Dielectric Spectroscopy Studies J. Non-Cryst. Solids 2015, 407, 478-485 10.1016/j.jnoncrysol.2014.08.057
- [45] Panagopoulou, A.; Kyritsis, A.; Aravantinou, A. M.; Nanopoulos, D.; Serra, R. S. I.; Ribelles, J. L. G.; Shinyashiki, N.; Pissis, P. Glass Transition and Dynamics in Lysozyme-Water Mixtures Over Wide Ranges of Composition Food Biophys. 2011, 6, 199-209 10.1007/s11483-010-9201-0
- [46] Panagopoulou, A.; Kyritsis, A.; Shinyashiki, N.; Pissis, P. Protein and Water Dynamics in Bovine Serum Albumin-Water Mixtures over Wide Ranges of Composition J. Phys. Chem. B 2012, 116, 4593-4602 10.1021/jp2105727
- [47] Khodadadi, S.; Curtis, J. E.; Sokolov, A. P. Nanosecond Relaxation Dynamics of Hydrated Proteins: Water versus Protein Contributions J. Phys. Chem. B 2011, 115, 6222-6226 10.1021/jp1122213
- [48] Shinyashiki, N.; Yamamoto, W.; Yokoyama, A.; Yoshinari, T.; Yagihara, S.; Kita, R.; Ngai, K. L.; Capaccioli, S. Glass Transitions in Aqueous Solutions of Protein (Bovine Serum Albumin) J. Phys. Chem. B 2009, 113, 14448-14456 10.1021/jp905511w
- [49] Johari, G. P.; Whalley, E. The Dielectric-Properties of Ice Ih in the Range 272-133-K J. Chem. Phys. 1981, 75, 1333-1340 10.1063/1.442139
- [50] Kurzweil-Segev, Y.; Greenbaum, A.; Popov, I.; Golodnitsky, D.; Feldman, Y. The Role of the Confined Water in the Dynamic Crossover of Hydrated Lysozyme Powders Phys. Chem. Chem. Phys. 2016, 18, 10992-10999 10.1039/C6CP01084A
- [51] Popov, I.; Puzenko, A.; Khamzin, A.; Feldman, Y. The Dynamic Crossover in Dielectric Relaxation Behavior of Ice I-h Phys. Chem. Chem. Phys. 2015, 17, 1489-1497 10.1039/C4CP04271A
- [52] von Hippel, A. The Dielectric-Relaxation Spectra of Water, Ice, and Aqueous-Solutions, and Their Interpretation. 3. Proton Organization and Proton-Transfer in Ice IEEE Trans. Electr. Insul. 1988, 23, 825-840 10.1109/14.8744
- [53] Bjerrum, N. Structure and Properties of Ice Science 1952, 115, 385-390 10.1126/science.115.2989.385
- [54] Sciortino, F.; Geiger, A.; Stanley, H. E. Effect of Defects on Molecular Mobility in Liquid Water Nature 1991, 354, 218-221 10.1038/354218a0
- [55] Sciortino, F.; Geiger, A.; Stanley, H. E. Network Defects and Molecular Mobility in Liquid Water J. Chem. Phys. 1992, 96, 3857-3865 10.1063/1.461890
- [56] Agmon, N. The Grotthuss Mechanism Chem. Phys. Lett. 1995, 244, 456-462 10.1016/0009-2614(95)00905-J
- [57] Fröhlich, H. Theory of Dielectrics: Dielectrics Constant and Dielectric Loss, 2 nd ed.; Clarendon Press: Oxford, 1986.
- [58] Geil, B.; Kirschgen, T. M.; Fujara, F. Mechanism of Proton Transport in Hexagonal Ice Phys. Rev. B 2005, 72 014304 10.1103/PhysRevB.72.014304
- [59] Bilgram, J. H.; Granicher, H. Defect Equilibria and Conduction Mechanisms in Ice Phys. Condens. Matter 1974, 18, 275-291 10.1007/BF01464397
- [60] Goto, K.; Hondoh, T.; Higashi, A. Determination of Diffusion-Coefficients of Self-Interstitials in Ice with a New Method of Observing Climb of Dislocations by X-Ray Topography Jpn. J. Appl. Phys., Part 1 1986, 25, 351-357 10.1143/JJAP.25.351
- [61] Hondoh, T.; Itoh, T.; Amakai, S.; Goto, K.; Higashi, A. Formation and Annihilation of Stacking-Faults in Pure Ice J. Phys. Chem. 1983, 87, 4040-4044 10.1021/j100244a008
- [62] Hondoh, T.; Itoh, T.; Higashi, A. Formation of Stacking-Faults in Pure Ice Single-Crystals by Cooling Jpn. J. Appl. Phys. 1981, 20, L737-L740 10.1143/JJAP.20.L737
- [63] Cowin, J. P.; Tsekouras, A. A.; Iedema, M. J.; Wu, K.; Ellison, G. B. Immobility of Protons in Ice From 30 to 190 K Nature 1999, 398, 405-407 10.1038/18848

- [64] Kim, S.; Park, E.; Kang, H. Segregation of Hydroxide Ions to an Ice Surface J. Chem. Phys. 2011, 135 074703 10.1063/1.3625435
- [65] Lee, C. W.; Lee, P. R.; Kang, H. Protons at Ice Surfaces Angew. Chem., Int. Ed. 2006, 45, 5529-5533 10.1002/anie.200601317
- [66] Moon, E. S.; Yoon, J.; Kang, H. Energy Barrier of Proton Transfer At Ice Surfaces J. Chem. Phys. 2010, 133 044709 10.1063/1.3457379
- [67] Park, K.; Lin, W.; Paesani, F. Fast and Slow Proton Transfer in Ice: The Role of the Quasi-Liquid Layer and Hydrogen-Bond Network J. Phys. Chem. B 2014, 118, 8081-8089 10.1021/jp501116d
- [68] Park, S. C.; Jung, K. H.; Kang, H. H/D Isotopic Exchange between Water Molecules at Ice Surfaces J. Chem. Phys. 2004, 121, 2765-2774 10.1063/1.1770548
- [69] Uras-Aytemiz, N.; Joyce, C.; Devlin, J. P. Protonic and Bjerrum Defect Activity Near the Surface of Ice at  $T < 145$  K J. Chem. Phys. 2001, 115, 9835-9842 10.1063/1.1414315
- [70] Migchelsen, C.; Berendsen, H. Proton Exchange and Molecular Orientation of Water in Hydrated Collagen Fibers. An NMR Study of H<sub>2</sub>O and D<sub>2</sub>O J. Chem. Phys. 1973, 59, 296-305 10.1063/1.1679805